REMARKS ON PROTEINS: SUMMARIZING STATEMENTS

ALBERT SZENT-GYÖRGYI
Institute for Muscle Research at the Marine Biological Laboratory, Woods Hole, Massachusetts

I want to make my remarks on proteins led by experimental work that is not my own but that of Drs. Gergely, Mihalyi, and Andrew Szent-Györgyi, who just happened to work in my laboratory.

To make myself clear I will have to go back a little in the history of myosin. Ten years ago the myosin molecule looked like a simple thin rod, and we were all happy because in those days, once one gave the relative over-all dimensions of a molecule one had said everything about a protein that had to be said. (Since then, Dr. Laki has cut the myosin molecule in two, showing that the old molecule was probably a dimer of a smaller monomer.) In the earlier days we had no doubt about the nature of contraction. Nobody doubted that it is some sort of folding. Whether the folding was random or occurred at only certain points with straight stretches between did not seem important. There was folding and I, myself, felt sure that it was a regular folding, and ATP, when producing contraction, acted on the points of the folding.

This model has since been complicated very greatly by the work of Gergely, Mihalyi, and Andrew Szent-Györgyi, which showed that the myosin molecule is built of three subunits shunted in series. There are two kinds of such subunits within one molecule. The one kind was called (because of the low sedimentation constant) "L-meromyosin" (L standing for light), and the other was called "H-meromyosin" (H standing for heavy). There are two L's and one H in one molecule. We

do not know their sequence but the most reasonable assumption would be that the H is in the middle.

There is little doubt that these units are really preformed. They are not just artifacts but they are really subunits that are there within the myosin molecule since the reactions and activities of the myosin molecule are shared between the two. It is only the H that combines with ATP and splits it, and we have reason to believe that the L is involved in contraction because its reactions show the same dependence on ionic concentrations as does muscular contraction.

I do not know the meaning of all this but I am sure it has one. The first remark I want to make about proteins is this: if we study a protein and try to find out its structure there is one question we mostly forget to ask and that we always should ask, namely, that about the meaning of the structure found. The essential question to me is not how a protein is built, but why nature has put those atoms together in that very specific order. What was the property nature wanted to achieve by putting that great number of atoms together in that very specific way?

Another remark I want to make about proteins more specifically is in connection with the distribution of ATPase activity. There is now a fairly good agreement that the energy of contraction comes from the ATP molecule and that the utilization of that energy is connected with the splitting of the ATP molecule in one way or another. If the L meromyosin is what contracts and produces work and it is the H that liberates the energy of ATP, then one has to suppose that the energy has to go, somehow, from the H to the L.

Here one is faced with a very basic problem of how energy moves the muscle. This is not a specific question limited to myosin but one of the most general biological problems. This problem is: how does energy drive the living machine? To my mind this is one of the most fundamental biological questions.

One could advance two different theories. Taking muscle as example of the living machine, one could picture the action of ATP as a local point action, producing a local change on the molecule by some classical chemical interaction. This is one possibility. We could say, for instance, that contraction comes about by points here and there, losing or increasing their charge on the myosin molecule. This then could produce some sort of a folding, a doubling-up and herewith shortening. So contraction could be explained, tentatively, by a purely local action of the ATP that would change something only at one single point in a reaction which could be described by classical chemical symbols, by letters and dashes between.

The other logical possibility would be to suppose that the energy is released from the ATP molecule in some active and mobile form capable of moving, and capable of diffusing through a system, and that it goes from the H-meromyosin to the L, producing changes there. At the moment, we cannot decide between the two possibilities outlined because we have too few data. I have spent the last 4 years exploring the second possibility and the more I see of it the more I begin to believe in it. There are observations that speak greatly in its favor. One such observation, for instance, is related to studies of the bacterial flagella (Bacterium Protei). Those flagella are 2 μ long; in atomic dimensions, this is miles and miles. There are reasons for believing that the energy that moves a flagellum is liberated at its base and starts up a wave of contraction, which then runs along this very long fiber that must work all along to drive the bacterium forward. These flagella are very thin — only 130 A in diameter. So they can consist of only a simple strand of protein fibers and there can be no tube in the middle to supply ATP or any other chemical energy source. We have to assume that the energy, produced at the base of flagella, moves along this fiber and is dissipated on its way as it does work.

This complicates our problem very much, because if we suppose some mobile form of energy we also have to suppose

some structure within the protein that conducts that energy. About 15 years ago, Dr. Laki and I speculated about this problem and came to the conclusion that proteins may be semiconductors. Since then, Dr. Gergely, with M. G. Evans, worked on this problem and found evidence for continuous energy bands in proteins, though the evidence was not quite conclusive. There are several possible ways to propagate energy through proteins but I do not want to go into them. Suffice it to say that in order to have a mobile form of energy, we would have to have some structure in the protein that can propagate that energy. Here I come back to my first remark because if there is such a structure, let's say, a conduction band, there must also be a very specific atomic configuration, and nature may have put those atoms together in that very specific way to achieve fusion of the energy levels of atoms to a band on a quantum mechanics basis (and nature seems to know a great deal of quantum mechanics).

This was the situation with muscle 15 years ago, but today the picture is still more complex, and this is due to the work of Andrew Szent-Györgyi. A decade or so ago I built a new thermodynamics of muscle, based on the assumption that the contractile matter consisted of very small subunits that acted independently from one another in an all-or-none equilibrium reaction. I was the only living creature in this world who believed in this theory. Andrew believed it half-heartedly. He said: "If there are really units as you suppose, also outside your head, then one should be able to find them."

So Andrew began to tease the myosin molecule and soon he found that under very specific conditions — at a special concentration and temperature — this protein goes to pieces if put in urea. It falls into very small pieces of equal size, of a molecular weight of 5000 g. So if the L-meromyosin has a molecular weight of 100,000 g, then it is built of twenty such subsubunits. The really dramatic feature of this discovery is that these units are held together by secondary forces only. If there ever was a "molecule" in biochemistry,

it is the myosin molecule, because it is there to produce tension and so has to withstand strain. Now it turned out that this molecule is not a molecule at all. It is a conglomeration, a regular heap of very small units held together only by secondary forces.

What does this mean? Let us consider first the mechanism of contraction. It probably means that contraction involves some rearrangement in the relative position of these very small units, "protomyosins." So if the energy of the ATP molecule has to move the myosin it has to do something to a greater number of such units and the forces holding them together. How it can do this we do not know since we do not know what contraction is. We do not have the least idea, and the more we know about muscle the less we understand it. If this goes on in the end we will know everything and understand nothing.

Of course, everybody has his own pet theory of contraction and his pet model, of which there is a great number now on the market. But I am afraid the situation is similar to that of the holy elephant that had ninety-nine names, the real one being the hundredth, known only to the elephant himself.

There are questions that come up in one's mind in connection with this structure of myosin. The characteristic of this molecule is that it is composed of small basic units of molecular weight 5000 g. My experience is that nature works with a few basic principles and not with exceptions. If we find something striking in one place then we usually find a basic law behind it, and later find the same law applied in other places over and over again.

So the experience with myosin might mean that the basic units of protein structure are of the order of magnitude of the protomyosins, but there is no proof for this. One may also think that myosin is an exception, nature having made it from small units because myosin is the only molecule that has to move; if it were built of long filaments, it could not move or change its shape. I am inclined to think the other

way; that is, to suppose that the architecture of myosin represents a basic principle that all proteins are constructed likewise. Myosin is an exception only in so far as its basic units are loosely connected to enable the whole protein to move. Insulin is also built of very similar units of molecular weight 6000 g and Dr. Waugh just told you in his very beautiful lecture how these can join to make a very specific and stable fiber. If this represents a basic blueprint, then protein synthesis is accessible to a new interpretation. The protein synthesis might go then in two steps, the first being the building of these little units and the second the putting of them together. They may get together spontaneously, as Dr. Waugh's insulin fibers, but they may also need an organizer for this act. The one of these functions may be performed by DNA and the other by RNA. Whatever the case may be, these observations on the structure of myosin pose new problems.

Keratin has also been decomposed into smaller units. If one is unable to decompose all other proteins into such small units, this will not take away the possibility that they have the same basic structure, only they may have their "protoproteins" held together by means of covalent bonds, no mobility being needed.

I would like to sum up my remarks on proteins by saying that we should not be content to ask questions about the structure of proteins but should inquire also into the deeper meaning of the structures found. Owing to its specific function, generation of motion, nature has endowed the myosin with specific qualities that open new ways for its analysis and that may possibly lead to new basic concepts about protein structure and function.